Interaction Between the 5-Hydroxytryptamine Transporter-Linked Polymorphic Region (*5-HTTLPR*) and Negative Life Events in Adolescent Heavy Drinking

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ABSTRACT. Objective: This study investigates whether the reciprocal associations between negative life events and drinking over time differ as a function of *5-HTTLPR* (5-hydroxytryptamine [serotonin] transporter-linked polymorphic region) genotype (i.e., candidate gene and environment interaction and correlation) using large and population-based prospective data from adolescents. **Method:** A total of 4,916 White adolescents in the United Kingdom (mean ages = 16, 17, and 18 years old over three assessment points; 47% female) were used. Tri-allelic *5-HTTLPR* genotype was assessed; negative life events were assessed at ages 16 and 17; and frequency of heavy drinking was assessed at ages 16, 17, and 18. Path analyses after controlling for covariate interactions and multigroup cross-lagged analyses were conducted. **Results:** The null findings of candidate gene and environment interaction and correlation were found in the path analyses controlling for covariate interactions, and they were replicated in the multigroup cross-lagged analyses. No moderation by *5-HTTLPR* in the association of negative life events at age 16 with heavy drinking at age 17 as well as no association of negative life events at age 17 with heavy drinking at age 18 were found. Also, the *5-HTTLPR* genotype did not moderate the association of heavy drinking at age 16 with negative life events at age 17. **Conclusions:** Using large prospective data, it appears that there is no evidence for *5-HTTLPR*-moderated drinking following experience of negative life events and no support for *5-HTTLPR*-moderated selection to negative life events among mid/late adolescents. This finding may inform developmental patterns in gene and environment interaction effects by showing that the effects are less pronounced in mid/late adolescence than in early adolescence. (*J. Stud. Alcohol Drugs, 81,* 566–574, 2020)

INDIVIDUALS MAY DRINK to cope with stressful experiences, and the stress-dampening effects of consuming alcohol may reinforce drinking behaviors over time (Sher, 1987). Genetic factors, specifically serotonin system associated with stress response (Oroszi & Goldman, 2004), may strengthen individuals' susceptibility to negative environmental influences (e.g., family conflict, academic failure). Recent candidate gene and environment interaction (cG × E) studies have suggested that the associations between negative life events and drinking behaviors may differ as a function of serotonin transporter genotype, 5-hydroxytryptamine transporter-linked polymorphic region (*5-HTTLPR;* Enoch, 2012). Genetically based selection into certain environments is referred to as "gene and environment correlation" (Plomin et al., 1977). The likelihood of experiencing negative life events (e.g., interpersonal problems) has been suggested to differ as a function of *5-HTTLPR* because of its association with neuroticism (Sen et al., 2004) and rumination (Clasen et al., 2011).

A number of studies have reported significant but mixed cG × E effects between *5-HTTLPR* genotype and negative life events on drinking behaviors (Covault et al., 2007; Kaufman et al., 2007; Kim et al., 2015; Kranzler et al., 2012; Laucht et al., 2009; Nilsson et al., 2005; Olsson et al., 2005). Initially, the *5-HTTLPR* short (*S*) allele was hypothesized to reduce transcriptional activity of the serotonin transporter compared with the long (*L*) allele (Heils et al., 1996). However, later research has suggested that *5-HTTLPR* may be tri-allelic, with low-activity alleles (*S′* = the *S* and *LG* alleles) demonstrating reduced transcriptional activity compared with the high-activity allele (*L′* = the *LA* allele; Hu et al., 2006). Two meta-analyses reported small but significant (odds ratios [OR] = 1.15−1.18) associations of the *5-HTTLPR* genotype with risk for alcohol dependence (Feinn et al., 2005; McHugh et al., 2010), and another review reported a null finding (Villalba et al., 2015).

The presence of a *5-HTTLPR* short or low-activity allele (*S* or *S′*) exacerbated the effects of family conflict (Kim et al., 2015) or general stressful life events (Covault et al., 2007; Kranzler et al., 2012) on drinking frequency and quantity among adolescents and young adults. In contrast, the presence of a long- or high-activity allele (*L* or *L′*) ex-

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acerbated the effects of poor parental attachment (Olsson et al., 2005) or stressful life events (Laucht et al., 2009) on binge drinking among young adults. Further, the genotype comprising one long and one short allele (i.e., *LS*) has been associated with increased risk for drinking in the presence of poor family relationships (Nilsson et al., 2005) or childhood maltreatment (Kaufman et al., 2007) among adolescents. Additional research found no differences in the association of stressful environments with alcoholism as a function of *5-HTTLPR* (Dick et al., 2007).

These mixed findings may be due to small sample sizes and/or covariate interactions. The majority of the cG × E studies involving *5-HTTLPR* have sample sizes ranging from 127 to 393 (Covault et al., 2007; Kaufman et al., 2007; Kranzler et al., 2012; Laucht et al., 2009; Nilsson et al., 2005). Due to the small main effects of candidate genes on behavior, large samples are needed to examine these associations and resolve such inconsistencies in existing research. A review of 103 cG × E studies (Duncan & Keller, 2011) demonstrated that, with an exception of very large cG × E effect sizes, at least 600 participants are needed to achieve 80% power. In addition to a large sample size, controlling for the associations of covariates (e.g., sex, socioeconomic status) with *5-HTTLPR* genotype or with negative life events is needed by adding Covariate × Gene and Covariate × Environment terms (Keller, 2014). Sex differences in genotype frequencies or the different exposure to negative life events as a function of economic status may confound potential gene and environment interaction effects. However, these confounding effects rarely have been statistically adjusted in cG × E studies involving *5-HTTLPR,* with a few exceptions (Cicchetti & Rogosch, 2014).

In addition to cG × E effects, gene and environment correlation (i.e., whether the likelihood that individuals will experience negative life events differs as a function of drinking behaviors and *5-HTTLPR* genotype) needs to be examined. Heavy drinking adolescents may be more likely to experience subsequent negative life events (e.g., alcohol-related accidents, academic failure, conflict with family members) (Chatterji & DeSimone, 2005). A cyclical pattern may emerge such that individuals exposed to negative life events may be more likely to drink, and, subsequently because of their alcohol use, may be more likely to experience negative events, which in turn may increase further drinking (O'Doherty, 1991). Heavy drinking adolescents with high genetic risks for certain behavioral or personality characteristics may seek out or provoke even more negative life events. For example, individuals with a genetic liability for neuroticism have been shown to be more likely to experience interpersonal stressful life events (Kendler et al., 2003), and heavy drinking may strengthen the association. However, no research has directly examined the moderating role of *5-HTTLPR* genotype in the effects of prior alcohol use on subsequent experiences of negative life events. Examining this gene and environment correlation will elucidate combined underpinnings of the *5-HTTLPR* genotype and drinking behavior under selection or provocation of stressful environments, which reflects a more realistic phenomenon.

The current study aimed to resolve mixed findings on *5-HTTLPR* and stress interaction effects by using a large sample, controlling for covariate interactions and testing for both gene and environment interaction and correlation. We investigated whether the reciprocal associations between negative life events and adolescent drinking over time differed as a function of *5-HTTLPR* genotype using population-based data from ages 16 through 18 years old. This study focused on mid- to late adolescence because previous *5-HTTLPR* × Stress studies extensively focused on childhood stressful life events. By focusing on the relatively understudied late adolescence period, we aimed to investigate whether gene and environmental influences differ over developmental stages. Specifically, we tested whether (a) the effects of negative life events on adolescent heavy drinking (i.e., gene and environment interaction) and (b) the effects of heavy drinking on the experience of negative life events (i.e., gene and environment correlation) differed as a function of *5-HTTLPR* genotype a using tri-allelic genotype categorization (i.e., *S′* vs. *L′* alleles).

Method

Participants and procedures

The data were derived from the Avon Longitudinal Study of Parents and Children (ALSPAC), an ongoing, population-based longitudinal study in the United Kingdom (Boyd et al., 2013; Fraser et al., 2013). In the Avon area of Great Britain, 14,541 pregnant women who had an expected delivery date between April 1, 1991, and December 31, 1992, were recruited. An additional 904 mother–child dyads were added through postnatal recruitment efforts. Data collection began in early pregnancy, and study offspring and their parents were followed up almost annually for 25 years through postal or online questionnaires and clinical assessment visits. Ethical approval for the study was obtained from the ALSPAC Ethics and Law Committee and the Local Research Ethics Committees. Please note that the study website contains details of all the data that are available through a fully searchable data dictionary and variable search tool: http://www.bristol.ac.uk/alspac/researchers/our-data.

For the current analyses, the data collected from children ages 16, 17, and 18 were used. The final sample included data from 4,916 adolescents after excluding 10,529 participants (68%). The samples' data were excluded because adolescents did not provide biological samples for genotyping, they had dropped out by the time of genotype collection, or their genotypes were not successfully analyzed for low DNA sample quality or other genotyping difficulties (n = 9,644).

Also, non-White adolescents (n = 192) or those who did not complete the race variable (n = 693) were excluded to control for potential confounding effects of race (i.e., *5-HTTLPR* genotype frequencies differ across races) (Hu et al., 2006).

Attrition analyses showed no significant differences between included (n = 4,916) and excluded (n = 10,529) adolescents on experience of negative life events at age 16, $t(4964) = -.30$, $p = .76$, and at age 17, $t(4584) = -.44$, $p = .66$. However, compared with included adolescents, those excluded were more likely to be male, $\chi^2(1) = 4.83$, $p = .03$, Cohen's $h = 0.69$, and to be higher social class, $\chi^2(1) = 40.50$, $p = .00$, Cohen's $h = 0.32$. Those excluded also reported lower frequencies of heavy drinking at ages 16 through 18, $t(2978–4790) = -2.38$ to -3.81, $p = .00$ to $.02$, Cohen's $d = 0.07$ to 0.14, than those included.

Measures

Demographics. Child sex (*male* = 1; *female* = 0) obtained from birth notification and maternal social class (measured as manual vs. nonmanual occupation; *manual occupation* = 1; *nonmanual occupation* = 0) was included as covariates, due to potential confounding effects on drinking behavior (Casswell et al., 2003; Wilsnack et al., 2009).

Serotonin transporter gene polymorphism (5-HTTLPR). Genotyping for *5-HTTLPR* was performed using procedures described elsewhere (Wendland et al., 2008). Observed triallelic genotypes included low-activity allele carriers ($L'L'$ = 1,153, 24%; $L'S'$ = 2,508, 51%) and noncarriers ($S'S'$ = 1,255, 25%). Tri-allelic genotypes were dichotomized for the current analyses into carriers of at least one *5-HTTLPR* low-activity allele (n = 3,661; 75%) or noncarriers (n = 1,255; 25%). We used the two-group classification because previous cG × E literature more frequently used a dichotomized categorization, as opposed to a three-group categorization; in addition, some fMRI/sMRI (functional magnetic resonance imaging/structural magnetic resonance imaging) studies showed the differences in emotion processing circuitry in subjects with one or two copies of *S* vs. *L*/*L* homozygotes (Canli et al., 2005; Furman et al., 2011; Pezawas et al., 2005). Allele frequency using the tri-allelic categorization was in Hardy–Weinberg equilibrium, $\chi^2(1) = 2.12$, $p = .15$. For ancillary analysis, we also used three-group genotype categorization ($S'S'$ vs. $L'S'$ vs. $L'L'$) to examine whether cG × E interaction results are robust. This three-group categorization has been used in a previous study investigating Stress × *5-HTTLPR* on emotional symptoms in ALSPAC samples (Araya et al., 2009).

Negative life events. Participants responded to 21 items assessing their experiences (0 = *no;* 1 = *yes*) and perceptions (0 = *neutral or positive;* 1 = *negative*) of major life events (e.g., moving to a new neighborhood/school, parents divorcing, poor academic performance, family dying, serious injury/illness, experiencing bullying) at ages 16 and 17; participants reported on major life events occurring during the last 4 years at age 16 and during the last year at age 17. Experience of major life events was not assessed at age 18. Of the 21 items, only events endorsed as negative were used to calculate participants' sum score. For current analysis, the sum score was used.

Frequency of heavy drinking. Typical frequency of heavy drinking was assessed at ages 16, 17, and 18. Specifically, participants responded to an item assessing how often they had six or more drinks on occasion using a Likert scale of 0 (*never*), 1 (*less than monthly*), 2 (*monthly*), and 3 (*weekly or more*). One unit of alcohol was presented to participants as 1/2-pint average strength beer/lager or one glass of wine or one single measure of spirit. We used the heavy drinking measure because heavy drinking among 15- to 16-year-old adolescents is prevalent in European countries (39%) (Hibell et al., 2012), which is approximately three times more than that of adolescents in the United States. The overall G × E and gene and environment correlation results with heavy drinking frequency measure were similar to those with alcohol frequency and quantity in our data set (results available on request).

Data analytic strategies

Descriptive analyses and attrition analyses were conducted in IBM SPSS Statistics for Windows, Version 19.0 (IBM Corp., Armonk, NY). Path analyses and multigroup cross-lagged analyses were conducted in Mplus, Version 7.4. Maximum likelihood estimation with robust standard errors was used to deal with the nonnormally distributed alcohol and negative life events variables. To accommodate missing data, a full-information maximum likelihood (FIML) procedure was used. Based on all available data, FIML has been shown to yield excellent estimates of a likelihood function of each individual, generate reasonable standard errors, and be robust to violations of normality and random missing data (Graham et al., 2003). Missing data on negative events at ages 16 and 17 were 2,389 and 2,491 respectively, and the missing data of heavy drinking at ages 16, 17, and 18 were 2,464, 2,689, and 3,340, respectively. Thus, in Mplus, estimator of maximum likelihood parameter estimates with robust standard errors (MLR) was used to obtain the robust estimates of population parameters with missing data using all available information.

As shown in Table 2, path analyses were conducted to include all two-way interaction terms of each covariate (i.e., sex and maternal social class) with predictors to account for potential confounding interaction effects (Keller, 2014). Regarding cG × E effects, as shown in Models 1 and 2 (first and second columns of data), *5-HTTLPR* Genotype × Sex, Negative Life Events × Sex, *5-HTTLPR* Genotype × Social Class, and Negative Life Events × Social Class were included as predictors in addition to gene and environment

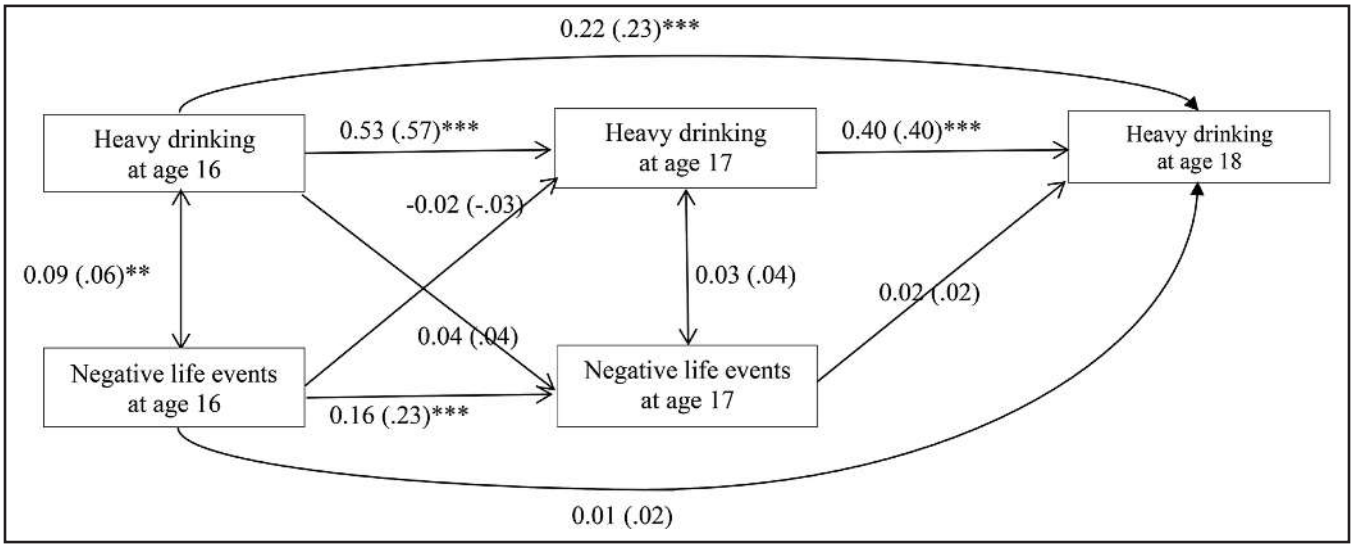

FIGURE 1. Unstandardized (outside parentheses) and standardized (inside parentheses) path coefficients resulting from the best-fitting final path models tri-allelic dichotomized categorization of the *5-HTTLPR* genotype on drinking frequency. In both models, sex and social class on all alcohol and negative life event variables at ages 16 through 18 were controlled for (covariate paths are not shown for simplicity).
$^{**}p < .01$; $^{***}p < .001$.

interactions on later alcohol outcomes. Regarding gene and environment correlation effect, as shown in Table 3, *5-HTTLPR* Genotype × Sex, Last-Year Heavy Drinking × Sex, *5-HTTLPR* Genotype × Social Class, and Last-Year Heavy Drinking × Social Class were included as predictors in addition to gene and heavy drinking interaction on later negative life events.

In addition, a multigroup cross-lagged model estimated the reciprocal associations between negative life events at ages 16 and 17 and alcohol frequencies at ages 16 through 18, as shown in Figure 1. Multigroup cross-lagged models do not allow us to control for covariate interactions such as genotype with covariate, but it estimates the reciprocal influence between negative life events and heavy drinking frequency over time. Auto-regressive paths represent changes within the same variable over time (e.g., heavy drinking at age 16 to heavy drinking at age 17). Contemporaneous paths represent cross-sectional associations between two different variables (e.g., association of heavy drinking at age 16 with negative life events at age 16). Cross-lagged paths represent prospective, reciprocal associations between different variables at different time points (e.g., heavy drinking at age 16 to negative life events at age 17). The main effect of sex on all study variables at all time points was controlled for in analyses (not shown in Figure 1 for simplicity). A 95% confidence interval (CI) using 2,000 bootstrap replications was reported.

To examine whether the reciprocal associations between alcohol use and negative life events differed as a function of *5-HTTLPR* genotype, we conducted multigroup analyses using a chi-square difference test. A chi-square difference test can determine whether there is a significant difference in model fit when making a comparison between a constrained (i.e., the path coefficient under examination is set to be the same across carriers of the low activity allele and noncarriers) and an unconstrained (i.e., each path coefficient under examination is free to vary across carriers and noncarriers) model. Chi-square difference tests were conducted individually for all auto-regressive, contemporaneous, and cross-lagged paths.

Last, the effect sizes of genetic differences in any reciprocal associations between alcohol and negative life events were estimated using Cohen's *d,* which is a standardized measure of the magnitude of difference between two means (Cohen, 1988).

Results

Descriptive statistics

Independent-sample *t*-tests and chi-square difference tests demonstrated no significant differences in any study variables as a function of *5-HTTLPR* genotype ($.24 < p < .99$; data available on request). Means (or percentages) of study variables in the entire sample, as well as their bivariate correlation coefficients, are shown in Table 1. Because of small mean values of the negative life events variable (M = 2.01 at age 16 and M = 1.07 at age 17) and little variability, the distribution graphs of negative events are presented in Supplemental Figure A. (Supplemental material appears as an online-only addendum to this article on the journal's website.) Spearman's correlation coefficient was reported due to nonnormality of distribution. Because of the nonnormality of the negative life event variables, we reviewed median in

Table 1. Means (and standard deviations in parentheses) and bivariate correlations of study variables

Variable	*M* (*SD*) or %	*r* 1.	2.	3.	4.	5.	6.	7.
1. Male sex	53%	–						
2. Maternal social class (manual occupation)	19%	.02	–					
3. Low activity allele	75%	.01	.01	–				
4. Negative life events at age 16	2.05 (1.56)	**-.17**	.01	.00	–			
5. Negative life events at age 17	1.07 (1.05)	**-.10**	.02	-.03	**.23**	–		
6. Heavy drinking frequency at age 16	1.21 (0.98)	.03	-.03	.01	**.05**	**.05**	–	
7. Heavy drinking frequency at age 17	1.35 (0.93)	**.09**	.04	-.01	-.01	**.06**	**.54**	–
8. Heavy drinking frequency at age18	1.81 (0.90)	**.06**	.02	.03	.03	**.06**	**.43**	**.51**

Notes: $N = 4{,}916$. Correlation coefficients significant at a p value of .05 are in **bold.**

addition to the mean values. The median value of negative events at age 16 is 2, and the median at age 17 is 1, which were not significantly different from mean values of 2.05 and 1.07, respectively.

Associations of the *5-HTTLPR* genotype with all study variables were not significant at p value of .05 (r = -.03 to .03). Males were more likely to report heavy drinking than females at age 17 (r = .09, p = .00) and at age 18 (r = .06, p = .02). Greater experience of negative life events was cross-sectionally associated with greater frequency of heavy drinking at both age 16 (p = .01) and age 17 (p = .02), but the magnitudes of the correlations were small (r = .05 to .06). Greater frequency of heavy drinking at age 16 was associated with greater experience of negative life events at age 17 (p = .03), but the correlation coefficient was also small (r = .05). In addition, the association graphs between the negative events and heavy drinking variables at different time points for the three-group genotype categorization are presented in Supplemental Figure B.

cG × E and gene and environment correlation after controlling for covariates' interactions

As shown in Table 2, path models after controlling for covariate interactions showed nonsignificant interaction between *5-HTTLPR* and negative life events at age 16 on frequency of heavy drinking at age 17 (b = -0.01; β = -.03, p = .60). No significant interaction effect between *5-HTTLPR* and negative life events at age 17 on heavy drinking at age 18 was found (b = 0.05; β = .06, p = .29). Regarding gene and environment correlation, as shown in Table 3, no significant interaction between *5-HTTLPR* and heavy drinking at age 16 on negative life events at age 17 was found (b = 0.06; β = .06, p = .38).

cG × E and gene and environment correlation in a cross-lagged model

Chi-square difference tests of all cross-lagged paths, as well as all auto-regressive and contemporaneous paths in the model, were not significant, $\Delta\chi^2(1) = 0$ to 2.26, p = .13 to 1.00. Accordingly, the best-fitting model (shown in Figure 1) had all paths constrained to be the same across genotype groups. This model showed an excellent fit to the data, $\chi^2(10) = 3.39$, p =.97; scaling correction factor = 1.02; root mean square error of approximation = 0.00; standardized root mean square residual = 0.01; comparative fit index = 1.00; and Tucker–Lewis index = 1.02. Regarding cG × E, there was no association between negative life events at age 16 and later frequency of heavy drinking at age 17 among both carriers and noncarriers of a low-activity allele (b = -.02, 95% bootstrapped CI with 2,000 replication samples [-0.04, 0.03]; β = -.03, p = .17). There was no association between negative life events at age 17 and later frequency of heavy drinking at age 18, among both carriers and noncarriers of a low-activity allele (b = .02, 95% bootstrapped CI with 2,000 replication samples [-0.02, 0.06]; β = .02, p = .40). Regarding gene and environment correlation, there was no association between frequency of heavy drinking at age 16 and later negative life events at age 17, among both carriers and noncarriers of a low-activity allele (b = .04, 95% bootstrapped CI with 2,000 replication samples [-0.03, 0.08]; β = .04, p = .12).

Effect sizes

Effect size analyses showed that the interaction effect between *5-HTTLPR* and negative life events at age 16 on frequency of heavy drinking at age 17 was very small (Cohen's d = 0.05). The effect size of the *5-HTTLPR* with negative life events at age 17 on heavy drinking at age 18 was small (Cohen's d = 0.13). The correlation effect between *5-HTTLPR* and heavy drinking at age 16 on negative life events at age 17 was also very small (Cohen's d = 0.07).

Ancillary analyses

There were no significant cG × E effects using three-group genotype categorization, thus corroborating the main finding with dichotomized genotype categorization. That is, chi-square difference tests of all cross-lagged paths, as well as all auto-regressive and contemporaneous paths in the

Table 2. Results from path models testing gene and environment interaction after controlling for covariates' interaction effects with predictors

Predictor	Outcomes: Model 1: Heavy drinking at age 17 β	Model 2: Heavy drinking at age 18 β
Male sex	.08	**.14**
Social class	.03	.06
5-HTTLPR (G)	.02	.04
Negative events at age 16 (E16)	-.02	.05
Negative events at age 17 (E17)	–	-.02
Heavy drinking at age 16 (Alc16)	**.56**	**.24**
Heavy drinking at age 17 (Alc17)	–	**.41**
G × E16	-.03	-.02
G × E17	–	.06
Covariate interactions		
G × Male	-.04	-.09
E16 × Male	.03	-.04
E17 × Male	–	.00
G × Social Class	-.02	-.04
E16 × Social Class	.00	-.01
E17 × Social Class	–	-.03

Notes: Correlation coefficients significant at a *p* value of .05 are in **bold.** G (gene) = *5-HTTLPR;* E (environment) = negative events; Alc (alcohol) = heavy drinking.

Table 3. Results from path models testing gene and environment correlation after controlling for covariates' interaction effects with predictors

Predictor	Outcome: Negative events at age 17 β
Male sex	-.06
Social class	-.07
5-HTTLPR (G)	-.05
Heavy drinking at age 16 (Alc16)	-.01
Negative events at age 16 (E16)	**.23**
G × Alc16	.06
Covariate interactions	
G × Male	-.01
Alc16 × Male	.01
G × Social Class	.05
Alc16 × Social Class	.09

Notes: Correlation coefficient significant at a *p* value of .05 is in **bold.** G (gene) = *5-HTTLPR;* Alc (alcohol) = heavy drinking; E (environment) = negative events.

model, were not significant, $\Delta\chi^2(2) = 0.11$ to 3.57, *p*s = .17 to .95. Also, as shown in Supplemental Table A, there were no significant differences in any study variables as a function of *5-HTTLPR* three-group categorization ($.34 < p < .85$). Last, we found no significant mediating role of depression in the pathway between genotype and drinking frequency (See Supplemental Figure C).

Discussion

We examined whether reciprocal associations between negative life events and adolescents' heavy drinking from ages 16 to 18 differed as a function of *5-HTTLPR* genotype. We examined both gene and environment interaction (cG × E) and correlation after controlling for covariate interactions with large prospective data. Results provide no evidence for genetically moderated drinking by *5-HTTLPR* in the presence of negative life events (cG × E). Also, results did not support *5-HTTLPR* genetically moderated selection to negative life events (gene and environment correlation) among adolescents who are frequently drinking heavily. These nonsignificant findings remained the same when sex and social class covariates' interactions were controlled for. The effect size analyses also showed very small to small gene and environment interaction effects (Cohen's $d = 0.05$–0.13) and correlation effects (Cohen's $d = 0.07$).

Our null finding is in line with those from other cG × E studies involving *5-HTTLPR* using large samples across diverse psychiatric outcomes. Several replication failures were reported in studies with large samples for depression and other outcomes ($n > 1{,}000$) (Chipman et al., 2007; Surtees et al., 2006), whereas significant cG × E effects have been mostly reported in studies with small samples (Duncan & Keller, 2011). The majority of studies finding significant cG × E effects between *5-HTTLPR* and negative life events on alcohol outcomes also have used relatively small samples ($n = 295$–393) (Covault et al., 2007; Kranzler et al., 2012; Laucht et al., 2009). One study using a large national sample ($n = 1{,}913$) found nonsignificant interaction effects (Dick et al., 2007).

In the cG × E field, there has been a particular concern about potential false positives because there are various environments that can be examined, and their definitions and measurements are diverse. There may be potential false negatives, because it is more difficult to statistically detect interaction effects compared with main effects (McClelland & Judd, 1993). Low statistical power may increase the possibility of both false positives and negatives, which impedes true discovery of cG × E effects (Duncan & Keller, 2011). This nonsignificant finding is in line with the lack of cG × E replicability to drinking reported in an emerging review (Pasman et al., 2019).

Although our previous study found significant Family Conflict × *5-HTTLPR* on drinking in early adolescence (Kim et al., 2015), the current study did not support an interaction effect of Negative Life Events × *5-HTTLPR* in mid-/late adolescence. Such findings might suggest developmental patterns in G × E effects. A large twin study (Kendler et al., 2011) revealed that cG × E effects on drinking behavior are much more pronounced in early adolescence than late adolescence and young adulthood because they are more malleable to environmental exposures at the early developmental stage; in other words, the genetic associations may be more plastic. Also, our prior study used a more specific, behaviorally operationalized measure of stress specific to the family environment. In contrast, the current study used a diverse measure of negative life events. In general, cG × E

studies with *5-HTTLPR* for depression examining specific and homogeneous stressors rather than the sum of negative events tend to show more consistent significant findings (for a review, see Caspi et al., 2010). The impact of specific family environment–related stressors and broader life event–related stress may differ from each other and need to be distinguished in cG × E studies. We selected this more diverse measure in an effort to capture wide-ranging environmental influences on drinking at this stage of development (peer, family, neighborhood) (Brown et al., 2008), compared with early adolescence when one's family environment is central. This finding suggests that *5-HTTLPR* might not interact with or have as strong an association with moderating response to diverse negative life events in mid-/late adolescence.

Regarding gene and environment correlation, we did not find support for such associations. Individuals carrying *5-HTTLPR* low activity (or short) allele and drinking frequently did not report more subsequent negative events than noncarriers. Nonsignificant correlation between *5-HTTLPR* genotype and negative life events also has been reported in previous research (Gillespie et al., 2005), although a number of twin studies demonstrated genetic factors associated with exposure to negative life events (Kendler & Baker, 2007). A review of previous gene and environment correlation studies across diverse mental illnesses (Jaffee & Price, 2007) indicated that gene and environment correlation can be more elaborately examined when behavioral or personality characteristics (e.g., aggressive disruptive behaviors, cognitive abilities, and temperament) are taken into account as mediators. Thus, future studies need to use a more sophisticated model to examine whether *5-HTTLPR* affects personality characteristics and increases risk for interpersonal negative life events.

Limitations of the current study and future directions are worth mentioning. First, the time references of negative life event measures at ages 16 and 17 were different, which may have yielded slightly distinct findings at the two time points. Specifically, adolescents reported experience of life events over the last 4 years at age 16 and life events over the last year at age 17. Given that the experience of negative events may change quickly (e.g., conflict with friends) or that the effects of negative events may be long standing (e.g., family conflict), these differences in assessments may have captured different life events at both ages.

Second, the variability of the two negative event variables was extremely minor, and 447 participants reported no negative events at age 16 and 803 reported no negative events at age 17, which may have yielded nonsignificant findings. Second, given that many participants were excluded from the final sample, the representativeness of the current sample may be limited. Thus, the current study result needs to be interpreted with caution, considering that included samples were more likely to be females, lower social class, and heavy drinkers than those excluded. Third, the current study only examined genetically based selection into negative life events at age 17, because we did not have the environment variable measured at age 18.

Future research should extend this gene and environment correlation investigation into adulthood. Gene and environment correlation effects may be more evident when individuals assume more active roles to construct their environments (Scarr & McCartney, 1983). Last, future studies may benefit from using polygenic approaches. Prior studies have suggested that individuals with high cumulative genetic risk scores of the five monoaminergic genotypes exposed to stressors have been found to be more sensitive to alcohol cues (Kim et al., 2019). A polygenic approach, including multiple genetic variants considering their combined impact, is necessary when investigating complex traits such as drinking behavior.

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Conflict-of-Interest Statement

The authors report no conflicts of interest.

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